In Vitro Forgiveness of Oral and Long-Acting INSTI-Containing Regimens at Drug Concentrations Simulating Variable Adherence

Rima K. Acosta, Michelle L. D'Antoni, Andrew Mulato, Stephen R. Yant, Tomas Cihlar, Kirsten White — Gilead Sciences, Inc., Foster City, California, USA

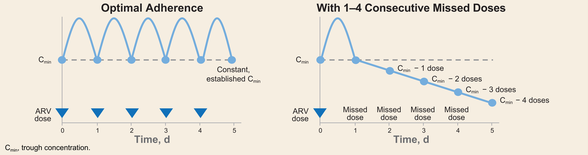
Gilead Sciences, Inc.
333 Lakeside Drive
Foster City, CA 94404
800-445-3235

Introduction

- EACS guidelines for initial treatment of HIV-1 infection recommend daily oral regimens anchored by an INSTI plus 1 or 2 NRTIs, including the STR of BIC/FTC/TAF, the combination of DTG+FTC/TAF, and the STR of DTG/3TC[1]
- The 2-drug daily oral STR of DTG/RPV is approved for PWH switching ARV regimens
- Recently, the LA INJ regimen CAB+RPV was approved for monthly and 2-month dosing in some countries
- Lapses in adherence to daily ARV drugs can lead to virologic failure and development of drug resistance, but regimens will have distinct durations of "forgiveness" (avoiding viral rebound and resistance in the setting of suboptimal adherence)
  - Long-acting regimens may be beneficial for those who want alternatives to daily oral medications, but they cannot be self-administered and there is potential for resistance development associated with low drug exposure, inconsistent dosing, preexisting drug resistance, or HIV-1 subtype[2]
  - Previous *in vitro* experiments have shown that when using an inoculum of wild-type or M184V virus, BIC+FTC+TAF was better at preventing viral breakthrough and emergent drug resistance than DTG+3TC[3]
    - *In vitro* viral breakthrough experiments should be analyzed comparatively; clinical trials assessing the impact of missed doses of these ARV combinations have not been conducted

3TC, lamivudine; ARV, antiretroviral; BIC, bictegravir; CAB, cabotegravir; DTG, dolutegravir; FTC, emtricitabine; INSTI, integrase strand transfer inhibitor; LA INJ, long-acting injectable; NNRTI, nonnucleoside reverse transcriptase inhibitor; NRTIs, nucleoside reverse transcriptase inhibitors; PWH, people with HIV; RPV, rilpivirine; STR, single-tablet regimen; TAF, tenofovir alafenamide.

Antiviral Pharmacokinetics of Daily Oral Regimens



C_{min}, trough concentration.

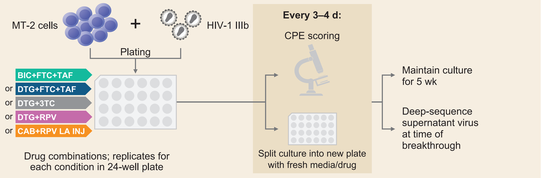
- Missing daily oral ARV doses results in a predictable decrease of systemic exposures to each drug in the regimen based on its established clinical half-life ($t_{1/2}$)

Objectives

- To understand relative time to *in vitro* viral breakthrough and resistance barrier using simulated human drug exposures at either full or suboptimal treatment adherence to BIC+FTC+TAF, DTG+FTC+TAF, DTG+3TC, DTG+RPV, and CAB+RPV LA INJ

Methods

Figure 1. *In Vitro* Viral Breakthrough Selections*



*MT-2 cells were infected with HIV-1 IIIb; infected cells were cultured in presence of fixed concentrations of BIC+FTC+TAF, DTG+FTC+TAF, DTG+3TC, DTG+RPV, and CAB+RPV, split every 3–4 d with fresh media containing drug, and monitored for viral breakthrough by cytopathic effect (CPE) for up to 5 wk; supernatants containing breakthrough virus were collected and stored.

- **Simulation of drug C_{min} *in vitro*:**
  - To simulate clinical C_{min}, pharmacokinetic data from participants in clinical trials were used and corrected for human plasma protein binding for BIC, DTG, RPV, and CAB
  - For the NRTIs FTC, 3TC, and TAF, intracellular active metabolite concentrations were used
- **Simulation of missed daily oral doses:**
  - To simulate 2 and 4 consecutive missed doses (C_{min} − 2 and C_{min} − 4, respectively), drug concentrations were adjusted by plasma $t_{1/2}$ for BIC, DTG, and RPV, and by intracellular $t_{1/2}$ for NRTIs (TAF, FTC, and 3TC)
  - C_{min} − X doses were determined as $C_{min} \times 0.5^{24 \times / t_{1/2}}$
- **Genotypic analyses:**
  - Each viral breakthrough supernatant was sequenced by next-generation sequencing (SEQ-IT GmbH & Co.KG, Kaiserslautern, Germany) and mutations were reported if present at ≥2%
    - A bioinformatics filter was used to remove APOBEC-mediated G-to-A hypermutated sequences
  - Mutations were observed between 2.1% and 69.3% per culture

Results

Table 1. Drug Concentrations for Cell Culture Equivalents

	Components of Daily Oral Regimens						LA INJ Regimen			
							q4wk (q1mo)		q8wk (q2mo)	
	BIC	FTC	TAF	DTG	3TC	RPV	CAB	RPV	CAB	RPV
Clinical dose, mg*	50	200	25	50	300	25	400	600	600	900
Molecular weight, g/mol	449.4	247.2	534.5	419.4	229.3	366.4				
Clinical C_{min}, µg/mL	2.61	0.096	0.008	1.11	0.042	0.08				
Clinical C_{min}, nM	5808	388	15	2515	265	218	6365	258	3602	176
Human serum shift[†]	43.6	1.0	1.0	27.5	1.0	32	74	32	74	32
$t_{1/2}$, h[‡]	17	37	116	14	17.5	50				
CCE C_{min}, nM[§]	133	388	15	91	265	6.8	86	8.1	49	5.5
C_{min} − 2	19	158	11	8.5	40	3.5				
C_{min} − 4	2.7	64.2	8.5	0.8	5.9	1.8				

*Clinical doses of BIC, FTC, and TAF in STR of B/F/TAF; DTG, 3TC, and RPV in STRs of DTG/3TC and DTG/RPV[4-6]; and CAB and RPV in CAB/RPV injectables[7]. †BIC, DTG and CAB data generated by standard equilibrium dialysis shift in human serum vs cell culture media[8]; RPV data generated internally and comparable to reported serum shift[9]. ‡Drug $t_{1/2}$ for BIC, DTG, FTC-triphosphate (FTC-TP), tenofovir-diphosphate (TFV-DP), 3TC-triphosphate (3TC-TP), and RPV[4,7,8,10-13]. §Cell culture equivalent (CCE) dose is clinical C_{min}/human serum shift ratio.

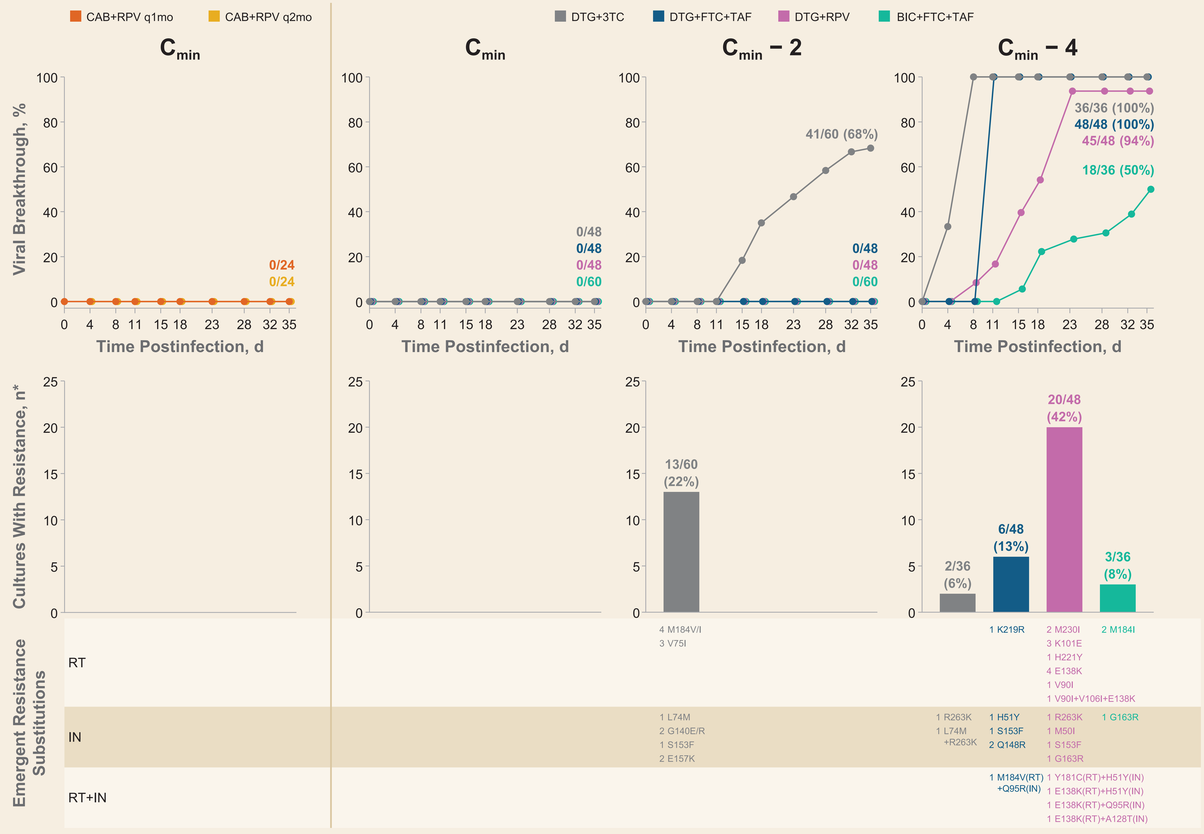
- BIC, DTG, RPV, and CAB concentrations were calculated using their human plasma clinical C_{min} according to their prescribing information, and adjusted for human plasma protein binding (Table 1)[4-10]
- TAF C_{min} used the active metabolite TFV-DP at its physiologic concentration in peripheral blood mononuclear cells from TAF-treated individuals[14,15]
- FTC and 3TC concentrations were set at their human plasma-free adjusted C_{min}[11-13]

Table 2. Mechanisms of Forgiveness and Barrier to Resistance for Daily Oral Combinations

	Mechanisms	BIC+FTC+TAF	DTG+FTC+TAF	DTG+3TC	DTG+RPV
Drug Levels	**Long plasma or intracellular $t_{1/2}$, h**				
	BIC or DTG	17	14	14	14
	FTC-TP or 3TC-TP	37	37	17.5	na
	TFV-DP	116	116	na	na
	RPV	na	na	na	50
	IN/DNA dissociation $t_{1/2}$ for BIC or DTG	132	71–78	71–78	71–78
Synergy and Mechanisms of Synergy	**Combination antiviral activity**				
	BIC or DTG + FTC or 3TC	Synergy	Synergy	Synergy	na
	BIC+TAF or DTG+TAF	Synergy	Synergy	na	na
	TFV+FTC	Synergy	Synergy	na	na
	DTG+RPV	na	na	na	Synergy
	TFV-chain-termination stabilized by dead-end complex with FTC-TP	Increased TFV activity	Increased TFV activity	na	na
Resistance Profile	**Phenotype of M184V**				
	BIC, DTG, or RPV	Sensitive	Sensitive	Sensitive	Sensitive
	FTC or 3TC	Resistant	Resistant	Resistant	na
	TFV (TAF)	Hypersensitive	Hypersensitive	na	na
Genetic Barrier to Resistance	**No. of mutations required to confer resistance**				
	BIC, DTG, or TAF	High	High	High	High
	RPV	na	na	na	Low–medium
	FTC or 3TC	Low	Low	Low	na

IN, integrase; na, not applicable.

Figure 2. Time to Viral Breakthrough



*Contained ≥1 known resistance-associated substitution. RT, reverse transcriptase.

- The LA INJ combination CAB+RPV prevented viral breakthrough at drug C_{min} (monthly and 2-month dosing; Fig 2)

Conclusions

- The INSTI-containing combinations of BIC+FTC+TAF, DTG+FTC+TAF, DTG+3TC, and DTG+RPV had no viral breakthrough with concentrations simulating high adherence
  - Regimen differentiation occurred when multiple missed doses were simulated *in vitro*; BIC+FTC+TAF had the highest forgiveness and barrier to resistance
    - *In vitro* viral breakthrough experiments should be analyzed comparatively; controlled clinical trials assessing the impact of missed doses of these ARV combinations have not been conducted
- The long-acting injectable combination of CAB+RPV had no viral breakthrough at concentrations simulating C_{min} for both monthly and 2-month dosing; further studies with CAB+RPV are needed to understand forgiveness during the pharmacokinetic tail of this regimen

References: 1. EACS. Guidelines Version 10.1; October 2020; **2.** Cutrell A, et al. AIDS 2021;35:1333-42; **3.** Mulato A, et al. JAIDS 2021;86:369-77; **4.** Biktarvy [package insert]. Foster City, CA: Gilead Sciences, Inc.; revised 9/21; **5.** Dovato [package insert]. Research Triangle Park, NC: ViiV Healthcare; 3/21; **6.** Juluca [package insert]. Research Triangle Park, NC: ViiV Healthcare; 3/21; **7.** Cabenuva [package insert]. Research Triangle Park, NC: ViiV Healthcare; 1/21; **8.** Margolis D, et al. Lancet 2017;390:1499-510; **9.** Tsiang M, et al. Antimicrob Agents Chemother 2016;60:7086-97; **10.** European Medicines Agency. CHMP assessment report: Edurant; 9/22/11; **11.** Dickinson L, et al. Antimicrob Agents Chemother 2015;5910:6080-6; **12.** Else LJ, et al. Antimicrob Agents Chemother 2011;56:1427-33; **13.** Yuen GJ, et al. Antimicrob Agents Chemother 2004;48:176-82; **14.** Callebaut C, et al, PLoS One 2017;12:e0169948; **15.** Custodio JM, et al. Antimicrob Agents Chemother 2016;60:5235-4. **Acknowledgments:** The authors thank Alex Thielen and Martin Daeumer at Seq-IT, and Ross Martin and Silvia Chang at Gilead for deep-sequence analyses. This study was funded by Gilead.